Centromere protein A dynamics in human pluripotent stem cell self-renewal, differentiation and DNA damage.

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Public Summary:
Human pluripotent stem cells (hPSCs) hold significant promise for use in regenerative medicine, or as a model to understand human embryo development. However, the basic mechanisms required for proliferation and self-renewal of hPSCs have not been fully uncovered. Proliferation in all eukaryotes is dependent upon highly regulated expression of the histone H3 variant Centromere protein A (CENP-A). In the current study, we demonstrate that hPSCs have a unique messenger ribonucleic acid (mRNA) reserve of CENP-A not found in somatic fibroblasts. Using short hairpin RNA technology to reduce but not ablate CENP-A, we show that CENP-A-depleted hPSCs are still capable of maintaining a functional centromeric mark, whereas fibroblasts are not. However, upon induction of differentiation or DNA damage, hPSCs with depleted CENP-A arrest in G2/M and undergo apoptosis. Analysis of CENP-A dynamics following DNA damage in hPSCs reveals that 60 min after irradiation, CENP-A is found in multiple small nuclear foci that are mutually exclusive to gammaH2AX as well as CENP-C. Furthermore, following irradiation, hPSCs with depleted CENP-A mount a normal apoptotic response at 6 h; however at 24 h, apoptosis is significantly increased in CENP-A-depleted hPSCs relative to control. Taken together, our results indicate that hPSCs exhibit a unique mechanism for maintaining genomic integrity by possessing the flexibility to reduce the amount of CENP-A required to maintain a functional centromere under self-renewing conditions, and maintaining a reserve of CENP-A mRNA to rebuild the centromere following differentiation or DNA damage.

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